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A COMPARISON OF THE EFFICIENCY OF METHODS OF DIAGNOSING EQUINE PREGNANCY, WITH SPECIAL REFERENCE TO THE MUCIN TEST

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INTRODUCTION.

It is of great importance to breeders of horses to be able to determine as early after service as possible which mares in a stud are pregnant and which are barren. If this knowledge is available before the end of the serving season, steps can be taken to have barren mares clinically examined and treated and thereby the loss of a breeding season in one or more animals may be avoided. Definite knowledge at the earliest stage possible enables the owner to make his arrangements to the best advantage. Pregnant mares can be turned off to grass, given suitable work and feeding, or otherwise dealt with, while barren mares can be got up for training, schooling or work, or may be sold, etc. The early subjective signs of pregnancy are, however, often misleading, and the breeder cannot rely upon sexual behaviour as an indication of pregnancy. It becomes necessary in consequence to have resort to certain tests or examinations for diagnosing pregnancy. Of recent years a number of these have been developed and it is of value to make a comparison between some of them.

METHODS OF DIAGNOSIS AVAILABLE.

Diagnosis of pregnancy can be attempted by (i) Clinical Methods, which include rectal palpation, vaginoscopy, and the mucin* test; (ii) Chemical Estimations, and (iii) Biological tests carried out on laboratory animals, the latter two groups depending upon the identification of the response to hormones present in blood serum or urine. Details of the techniques employed for the biological determination of oestrogens in urine, and of gonadotrophic substances in blood serum are now standard practice and need not be referred to further. The technique most conveniently employed for carrying out rectal examination has been described in some detail (Miller and Day, 1939) and may be consulted in this connection, but observations made at definite intervals during pregnancy are included later.

The Mucin Test

The technique of the *Mucin Test* is not so well recognised and a description is included for readers' convenience.

A speculum (Polansky's as modified by Messrs. Arnold is perhaps most convenient) is introduced into the vagina, and by the use of a head-lamp, or a narrow long-handled torch, vaginoscopy is carried out. The cervix is carefully inspected as well as the walls of the vagina. A dry long-handled (20 in.) paint brush is then introduced and the bristles gently rotated over the os of the cervix, so that a sample of the mucus present adheres to the brush. The brush is withdrawn and the mucoid secretion is spread over the surface of a clean microscopic slide. The consistency of the mucus in the vagina is noted. The slide is subsequently fixed with alcohol, dried, and stained for 20 minutes with Delafield's haematoxylin, or for one minute in 1 per cent. aqueous methylene blue, washed off with tap water (in each case), dried and examined.

CHANGES IN VAGINA AND CERVIX DURING OESTRUS, DIOESTRUS AND PREGNANCY. I. *Oestrus.*—In normal mares the mucosa of vagina and cervix varies from a bright pink to a light red in colour. The surface is moist and glistening due to the fluid nature of the mucoid secretion; this is spread evenly over the surface, is plentiful and has an appearance similar to saliva. The cervix is relaxed and the mucosal folds in the cervix generally have an oedematous appearance.

II. *Dioestrus.*—The appearance of the vaginal and cervical mucosa is pale pink or slightly blanched pink in colour, and dry-looking instead of moist and glistening. The cervix is constricted, erect, and the os is plainly visible at its apex, where the mucosal folds meet. In position it is almost invariably in the mid-line and projects directly backwards. The mucoid secretion is very scanty, but is slightly sticky and tenacious to the touch.

III. *Early Pregnancy.*—In some mares commencing as early as the 25th day after service, in others later, the vagina and cervix become very pale, blanched and show a pearly appearance. Small blood vessels and capillaries are seen prominently as a network over the surface. As pregnancy advances these disappear and the whole of the mucous membrane has a bloodless or blanched appearance. The folds of the cervix become sealed together by the thick, tacky mucoid secretion, so that the external os seems to be completely obliterated. The apex of the cervix is normally turned away from the mid-line, generally downwards and to one side, but sometimes upwards.

The arms of the closed speculum do not slip in readily and the vaginal walls do not balloon as in the barren mare, but separation of the walls occurs slowly as though they were peeling apart from each other with

*Throughout this paper, the word " mucin " is used to indicate the test which depends upon the presence of a mucoid secretion in the cervix and anterior vagina. It has not been determined whether this material is actually mucin or whether it is a muco-protein.

some little difficulty. The whole mucosa has a very dry appearance and its secretion is sticky or even gum-like (according to the stage of pregnancy) and when spread on the slide resembles honey. The amount and the stickiness of the secretion in the anterior vagina increases with the advance of pregnancy.

Examination of Slides.—In stained smears from pregnant mares, affinity for stains is very marked. They are thick (it is almost impossible to spread the secretion thinly and evenly), and dark in colour, whereas in the barren mare the mucous material is thin and the smear appears pale on the slide. The pregnant smears under 2/3 objective show distinct globules of mucus (probably mucin or a muco-protein) and it is always possible to find large numbers of cells. Under 1/6 lens, the co-called pregnancy cells can usually be identified. These are columnar epithelial cells, produced by the cervix, which become entangled and accumulate in the thick tenacious secretion. They show various forms, commonly having the appearance of a bent tin-tack, with a dark-staining nucleus near their points. The cytoplasm of the cells stains faintly so that the cell may have a "ghost" outline on a background of more densely stained secretion. With methylene blue the dense blue background may mask entirely the presence of the cells, but this is rare with the haematoxylin, which has an advantage in consequence, though the time needed for staining is longer.

In barren mares, no globules of mucous secretion are found, there are few columnar epithelial cells, or none may be found, but there are usually polymorphs present, and there may be a variable number of cells of other kinds present as well.

The diagnosis of pregnancy is based upon finding globules of mucous secretion, numbers of "pregnancy cells," and perhaps a small proportion of other cells. The thickness and intensity of staining of the smear are also taken into account.

Results

The following tables have been drawn up to give the comparative results obtained in 1937 and 1938 from a certain number of selected mares examined at different stages of pregnancy. A few mares were examined during both years, and enter into both Tables I and II, but most of the mares were examined during one season only. The urine tests carried out are not really large enough to permit of percentages being given from which the accuracy of this particular test can be judged, and this remark applied equally to the number of blood serum examinations made during 1938. The figures are however included for convenience of reference. It should perhaps be emphasised that the level of excretion of oestrogens in mare's urine varies markedly in the earlier stages of pregnancy in many animals. The amount excreted may change considerably from day to day until the end of the first third of the gestation period (110 days approx.); thereafter it becomes more regular and increases steadily during the second third of the pregnant phase (up to about 250 days or so) to fall again to lower levels during the last 3½ months before foaling occurs. This and other related phenomena will be referred to further in the discussion.

Table I

Season 1937. *Number of Mares Examined for Each Individual Test and Percentage Accuracy of Test*

Stage of Pregnancy in Days.	Blood Test.			Mucin Test.			Urine Test.		
	No. Correct.	No. Wrong.	Percentage Accuracy.	No. Correct.	No. Wrong.	Percentage Accuracy.	No. Correct.	No. Wrong.	Percentage Accuracy.
20–39 ...	18	19	48·6	52	12	**81·5**	15	16	48·4
40–49 ...	19	3	**86·4**	31	7	**81·6**	7	10	41·2
50–59 ...	26	4	**86·7**	35	7	**83·3**	6	12	33·3
60–69 ...	23	1	**95·8**	27	5	**84·4**	3	4	42·8
70–79 ...	25	2	**92·6**	33	2	**94·3**	4	7	36·4
80–89 ...	19	4	**82·6**	26	3	**89·6**	5	3	**62·5**
90–99 ...	24	4	**85·7**	31	1	**96·9**	3	13	**18·8**
100–120 ...	29	9	**76·3**	50	4	**92·6**	8	7	**53·3**
Over 120 ...	18	18	50	60	2	**96·8**	10	1	**90·9**

Table II

Season 1938. *Number of Mares Examined for Each Individual Test and Percentage Accuracy of Test*

Stage of Pregnancy in Days.	Blood Test.			Mucin Test.			Urine Test.		
	No. Correct.	No. Wrong.	Percentage Accuracy.	No. Correct.	No. Wrong.	Percentage Accuracy.	No. Correct.	No. Wrong.	Percentage Accuracy.
20–39 ...	3	4	42·9	31	12	**72·1**	2	3	40·0
40–49 ...	11	3	**78·6**	23	7	**76·7**	1	2	33·3
50–59 ...	13	3	**81·3**	31	5	**86·1**	2	3	40·0
60–69 ...	11	0	**100·0**	29	7	**80·1**	2	3	40·0
70–79 ...	15	1	**93·8**	42	2	**91·3**	2	5	28·6
80–89 ...	9	1	**90·0**	28	2	**93·3**	5	2	**71·4**
90–99 ...	7	1	**87·5**	30	1	**96·8**	4	2	**66·6**
100–120 ...	13	1	**92·9**	40	1	**97·6**	9	1	**90·0**
Over 120 ...	11	9	55·0	81	3	**96·4**	19	6	**76·0**

TABLE III

Seasons 1937 *and* 1938. *Number of Mares Examined by Each Test and Percentage Accuracy of Results Obtained.*

Stage of Pregnancy in Days.	Blood Test.			Mucin Test.			Urine Test.		
	No. Correct.	No. Wrong.	Percentage. Accuracy.	No. Correct.	No. Wrong.	Percentage Accuracy.	No. Correct.	No. Wrong.	Percentage Accuracy.
20–39 ...	21	23	47·7	83	24	77·6	17	19	47·2
40–49 ...	30	6	83·3	54	14	79·4	8	12	40·0
50–59 ...	39	7	84·8	66	12	84·6	8	15	34·7
60–69 ...	34	1	97·1	56	12	82·4	5	7	41·7
70–79 ...	40	3	93·0	75	4	94·9	6	12	33·3
80–89 ...	28	5	84·8	54	5	91·5	10	5	66·6
90–99 ...	31	5	86·1	62	2	96·9	7	15	33·6
100–120 ...	42	10	80·8	90	5	94·7	17	8	68·0
Over 120 ...	29	27	51·8	141	5	96·6	29	7	80·5

In Table III are given the combined results for both years. These resemble the results obtained in individual years, shown in Tables I and II.

It is of some importance in considering the foregoing results to know in each case whether the incorrect results obtained were barren mares which were diagnosed as pregnant, or *vice versa*. In the following table (IV), which gives the combined results for both seasons, "wrong positive" means that the test indicated that the mare was pregnant (*i.e.*, positive) whereas she proved later to be barren; similarly a "wrong negative" is a pregnant mare which gave a negative result to the particular test.

TABLE IV

Combined Results for 1937 *and* 1938

Test.	Number of Mares Examined.	Total Number of Examinations.	Number of Wrong Positive Results.	Number of Wrong Negative Results.
Mucin	545	765	59	20
Blood serum ...	180	381	8	26
Urine	101	207	0	43

It is perhaps noteworthy that in the mucin test pregnant mares were diagnosed incorrectly three times as often as barren mares; in the blood test barren mares were wrongly diagnosed about three times more often than pregnant mares; while in the urine test all the errors were in mares which were actually pregnant at the time but gave negative results at the test. This latter fact is concerned mainly with the excretion of oestrogens in the urine not reaching test threshold levels until a later stage of gestation.

In Table IV only the results which were incorrect for the period when each test is normally applicable to the mare have been included; *i.e.* errors in the blood test between 42 and 120 days after service; in the urine test after the 85th day, and in the mucin test, after the 21st day.

A certain number of mares were examined more than once and a single mare sometimes contributed two or more errors in diagnosis at subsequent examinations.

From these results it will be noted that the tests on urine for oestrogens gave the least satisfactory results. This can be ascribed to two different causes. First, owing to the receipt of large numbers of samples during an exceedingly busy period, some of the samples had to be kept in a refrigerator for from one and a half to four months. It is very likely that during this time, when bacteriological activity could not be completely controlled, some breakdown or change in the molecules of the oestrogens occurred, whereby their capacity to provoke a response in the test animals was lost or diminished. In the second place (see Tables I to III), it will be noted that in the earlier stages of pregnancy the number of correct results is less than the number of errors, so that percentage of accuracy is less than 50 per cent. Diagnosis of pregnancy based upon chance selection alone should give results which are approximately 50 per cent. right and 50 per cent wrong, provided a large enough population of mares is taken. The results suggest that the number of examinations made was not large enough.

There are, however, some pregnant mares in which excretion of oestrogens is extremely variable at different times. No explanation can be offered which will account for such variations as those which occurred in one particular group of eight mares, which were included in the results for 1937-38 (Tables I to III). These were heavy draught mares all kept at one farm and under conditions which were essentially identical, so far as feeding, management, etc., are concerned. Table V (overleaf) indicates the results which were obtained.

The second examination was carried out 101 days after the first. In the intervening period opportunity occurred during five weeks to have assays made upon the bulked urine from the eight mares for oestrone content. This was actually concerned with an independent investigation, but the results are sufficiently interesting to be worthy of inclusion here.

The urine was collected daily as opportunity arose and a mixed week's supply was sampled for the purposes of the determination. Each week's supply was treated in the same way in all respects. The

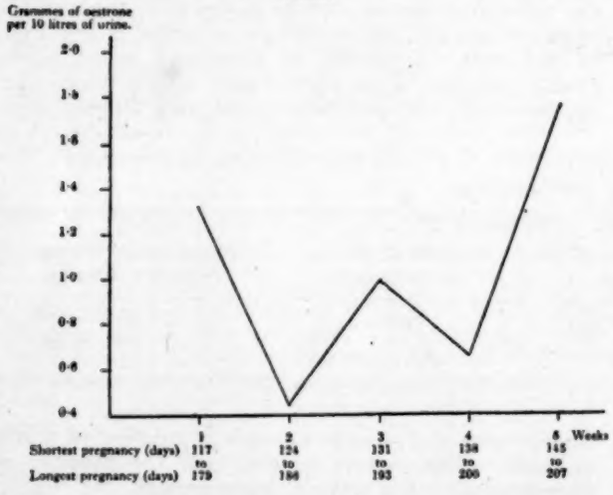
TABLE V

Designation of Mare.	1st Examination.				2nd Examination.		
	Stage of Pregnancy. in Days.	Results of Tests Obtained.			Stage of Pregnancy in Days.	Result of Urine Test.	Time at which Rectal Examination Confirmed Results (days).
		Blood.	Urine.	Mucin.			
A	128	—	—	+	229	+	210
B	150*	—	—	+	251	+	232
C	130	+		+	231	+	212
D	140	—	—	+	241	+	222
E	95	—	—	+	196	+	177
F	88*	+	+	+	189	+	170
G	104	—	—	+	205	+	186
H	150	—	—	+	251	+	232

* Note.—The difference between these—the extremes—is 62 days.

results are perhaps best shown in graphic form. They are given in Fig. 1, the yield being expressed as grammes of oestrone per 10 litres of mixed urine.

Variation in excretion of oestrone by a group of eight mares. The mares were all pregnant and kept under the same conditions of feeding and management. The ranges of pregnancies of the mares in days are given for each week.



The low yields obtained at the second and fourth week, from mares between 124 and 186 days of pregnancy in the one case, and between 138 and 200 days in the other, suggest that some common factor affecting all the mares must have been in operation. We are not prepared to do more than suggest that it may possibly have been associated with temperature and intake of water. In cold weather the excretion of water by the kidneys is increased, and there is a correspondingly greater degree of dilution of each of the substances present in urine.

Some Errors.

Other types of error encountered are worthy of brief note. Correlation of mistakes, where two or more tests agreed in result but were incorrect, were few. In one mare, mucin and blood tests indicated pregnancy at 38 and 88 days, but the mare proved to be barren. In another mare, a mucin test at 35 days, mucin and blood test at 57 days and again at 81 days and a mucin test at 97 days, all gave positive results, but subsequently the mare proved to be barren. Another mare ultimately shown to be barren gave positive mucin and blood tests at 50 days, and later negative results, while in another case wrong positive results were obtained with blood and mucin tests at 59 days. In these four mares, we regard it as highly probable that at the time the examinations were carried out the mares were actually pregnant, but that at some time subsequently abortion or foetal resorption occurred unknown to owners or attendants.

Several mares, included in the group of eight to which reference has been made, gave incorrect negative blood and urine tests (see Table V).

During the 1939 season, after the experimental work was completed, a large number of mares were examined by one of us (F. T. D.) by the mucin test only. A few instances where this test gave erroneous results were encountered, but the great majority gave reliable results throughout the early and middle stages of pregnancy.

Errors Due to Vaginal Aspiration of Air.—In thoroughbred brood mares, intake of air into the vagina, caused either by injuries to the vulva at a previous foaling or by abnormal flaccidity of the vulva, is not uncommon. The condition occurs in other breeds but to a somewhat less extent. If it is severe, breeding may be impossible and the mare becomes sterile until surgical correction has been carried out. In less pronounced cases, mares may become pregnant, but the typical changes in the vagina and cervix do not become established until later than normal. Such mares up to 60 days or later may consequently give negative results to the mucin test, which is, of course, based essentially upon *normal* physiological changes.

Significance of Anoestrus. — Anoestrus normally begins about the month of October, and when barren mares have passed into this phase, a certain number may be diagnosed by the mucin test as pregnant since the vaginal and cervical changes are essentially the same in anoestrus and in pregnancy. The blood or urine tests (if the duration of expected pregnancy is appropriate) may give further help, or rectal examination may be carried out.

Cystic Ovaries in mares may give rise to errors in the mucin test, since a proportion of mares so affected will give the typical smear pictures of pregnancy, and the appearance of cervix and vagina may seem to confirm these. The number of such mares is however small and assistance in diagnosis can in most cases be obtained from the mare's history and behaviour, while rectal examination will reveal the presence of the diseased ovaries, and the absence of a foetus.

Appropriate Stages of Pregnancy for Each Test.

The conclusion reached from the work carried out is that none of the three tests is infallible, but that each can give valuable information when performed at the stage of pregnancy for which it is most appropriately suited.

The Blood Serum Test will give positive results in pregnant mares between the 40th and 120th days in the great majority of cases. The time of greatest concentration of gonadotrophic hormone in the circulating blood is between the 60th and 75th days. Thereafter the level falls until by the 120th day (or better by the 100th day) the test becomes inappropriate.

The Mucin Test gives positive results as early as 20 days after service, and with experience may be employed from the end of the fourth week of pregnancy. Its accuracy increases as pregnancy advances, being most effective after the 80th day. As indicated earlier, it has been extensively employed during the past three seasons for mares of many different breeds with a great measure of success.

The Urine Test is not applicable in the early stage of pregnancy since oestrogens are not normally being excreted in sufficient amounts. About 80 per cent. of pregnant mares should give satisfactory positive results at the 90- to 100-day stage, and many some 20 or 30 days sooner. It is however in the period succeeding the 120th day and up to about the 250th day that it is most useful. The results of the urine tests included earlier are not a true index of the usefulness of the test for reasons already given. Other experience of one of us (Wm. C. M.) in a larger number of mares may serve to give a more accurate estimate. The results in Table VI are taken in sequence (for each group or stage of pregnancy) from the records of testing a series of samples received from veterinarians and owners, who furnished the information regarding the mares. They include mares tested between 1935 and 1939.

Table VI

Results of Routine Tests on Samples of Urine

Stages of Pregnancy (days).	Number of Tests Carried Out.	Number of Incorrect Results.	Percentage of Error.
80–89 ...	85	10	11·7
90–99 ...	79	4	5·0
100–119 ...	131	11	8·4
120 and over	219	19	8·7
General average	514	44	9·3 per cent.

Rectal Palpation.

It is considered desirable to include observations upon rectal palpation, which have been accumulated as the result of an opportunity which presented for one of us (F. T. D.) to examine a batch of seven pony mares every few days from the date of service until foetuses could be readily distinguished. In one mare, pregnancy was determined on the 16th day and in all by the 23rd day, the stage of pregnancy being estimated not from the date of service but from the time of ovulation which was about two days later.

Before the foetus could be palpated, it was possible to anticipate whether any mare was pregnant by reference to the tone of the uterus on the 15th day. In barren mares it was soft and flaccid, like a fold of velvet fabric, in pregnant mares it was turgid and well defined, like a rubber sponge roll.

At the 20th day, the embryonic mass was the size of a bantam's egg, and in the early stages is best described as feeling similar to a " shell-less " (or soft-shelled) hen's egg.

By the 30th day, pregnancy can be diagnosed by palpation in most primiparous mares and in those which had no foal in the preceding season ; while by the 45th day it should be possible with adequate experience to diagnose pregnancy in the majority of normal mares.

As a guide to clinicians examining mares during the earlier stages of the first third of pregnancy, it may be useful to include Table VII which gives an approximate comparison between the size of the embryonic mass, membranes and fluid, lying in the uterus, and some common objects. This table is based upon work done on Welsh pony mares. For thoroughbred or larger mares the sizes of the embryonic masses are relatively larger at the given stages of pregnancy.

Table VII

Duration of Pregnancy in Days.	Size of Palpable Embryonic Mass.	Position in Uterus.
16	Bantam's egg	In uterine horn (often the right) beyond junction between body and uterine horns.
20	Golf ball	" "
25	Pheasant's egg	" "
30	Hen's egg	" "
35	Tennis ball	" "
40	Turkey's egg	" "
45	Goose egg	" "
50	Jaffa orange	Occupying 2/3 uterine horn and about 1/3 body.
60	Large Jaffa orange	" "
70	Ostrich egg	Occupying uterine horn and about 1/2 body of uterus.

It should perhaps be noted that prior to the 45th day of pregnancy, it may be difficult or impossible to palpate the embryonic mass in the uterus of some mares, but with care and experience it should usually be possible subsequent to this time.

The details of technique and results obtained by rectal examination of mares in the later stages of pregnancy have already been described (Miller and Day, 1939). It may perhaps be emphasised that all mares which are encountered by the practitioner will not necessarily present the same picture, since there are minor differential rates of development in some while needful allowances must be made for mares of different sizes, breeds, etc. It will be found, however, that the great majority will be found to fall within the framework of the above suggestions.

Summary

The paper gives an account of the usefulness of diagnosing pregnancy in mares by mucin test and by examinations of blood and urine. The results which have been obtained are given, and it is indicated that

while no one test is infallible, each has its uses. The mucin test has the widest range of application. Its technique is described in detail.

Blood serum examination for presence of gonadotrophic hormone is useful between the 40th and 120th days after service; urine examination for oestrogens is useful after the 120th day and until towards the end of pregnancy. The reasons for erroneous diagnosis based upon the results of these examinations are briefly discussed.

The use of rectal examination as a means of diagnosis is considered, and a table is included showing a comparison between the size of the embryonic mass and common objects for the earlier stages of pregnancy.

REFERENCE

MILLER, WM. C., and DAY, F. T. (1939.) The Diagnosis of Equine Pregnancy. *J. roy. Army Vet. Corps.* May, 1939. pp. 95-105.

TRANSMISSION OF TUBERCULOSIS FROM MAN TO CATTLE

"The transmission of tuberculosis from cattle to man is so much more important than the reverse that the possibility of dairy cattle being infected by their attendants will be a new conception to many," comments the *Lancet* in a recent in annotation. "It is true that the Public Health (Prevention of Tuberculosis) Regulations of 1925 prohibit the employment in a dairy of persons suffering from respiratory tuberculosis, but this clause was probably intended primarily to guard against direct infection of the milk. Nielsen and Plum[1] of the State Veterinary Serum Laboratory of Copenhagen have now recorded instances of infection of cattle by tuberculous persons employed on farms in Denmark. Farms which had been free from tuberculin reactors revealed without warning a surprising number of positive reactors, suggesting the sudden appearance of an unexpected focus of infection in their midst. To give one instance, a herd which had been free from tuberculosis since 1932 suddenly yielded 19 positive reactors out of 29 cattle. On investigation a farm hand was discovered who had been previously vigorous and apparently healthy, but who was later admitted to hospital and subsequently transferred to a sanatorium. The records in the other farms quoted in the paper are similar. There were 17 farms in all, and in 13 of these a worker suffering from tuberculosis was found from whom the bovine strain of the tubercle bacillus was isolated. In one instance, a human-type strain was obtained from a worker, in two others the strain had not been typed, and in the remaining case the strain though bovine might conceivably have reached the herd during transport. Reid[2] has also recorded an instance in this country of a heifer infected with the human type of bacillus and cites two more from the literature published by A. S. Griffith in 1913. In only one of these three, however, was contact with a phthisical person established—one of Griffith's cases. Griffith[3] has discussed the question of cattle contacts from the point of view of direct transmission of bovine tubercle bacilli from cattle to man by contact. This possibility having now been well established there seems no inherent reason why the reverse process should not occasionally take place."

(1) NIELSEN, F. W., and PLUM, N. *Vet. J.* January, 1940, p. 6.
(2) REID, J. *J. comp. Path. Therap.* (1939.) 52. 318.
(3) *Tubercle.* (1936-37.) 18. 529.

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A report from Malmö states that an estate in the province of Skaane has been declared free from foot-and-mouth disease. This was the last case in Sweden, discovered on May 13th last, so that the country is now entirely cleared.

THE VETERINARY RECORD

OCTOBER 5TH, 1940

THE CHARITIES OF THE PROFESSION IN WAR-TIME

AN URGENT APPEAL TO ALL MEMBERS OF THE N.V.M.A.

DURING recent days every member of the Association has, or should have, received a copy of an appeal on behalf of the charities of the profession addressed by the officers to all those who, in large or small measure, can as a matter of extreme urgency, do what lies in their power to assist the benevolent societies in meeting the immediate or prospective drain on their slender financial resources arising from the demands of the war situation. Unfortunately, through a lapse in despatch, for which "emergency measures" were responsible, the appeal was sent out with the annual report in an open buff envelope which gave to this important communication also the unimpressive aspect of a circular, with the unhappy consequence that many members may have omitted to read it. In any event, in view of the supreme importance of a generous and prompt response to the welfare of many hard-hit veterinarians and their dependants, we make no apology for using this space and opportunity to impress the message and its moving implications, more deeply upon the minds of members who are in a position to help those of their colleagues who, through factors the effects of which no foresight on their part could avoid, now find themselves, in not a few instances, in truly desperate straits.

The text of the appeal is as follows:—

NATIONAL VETERINARY MEDICAL ASSOCIATION OF GREAT BRITAIN AND IRELAND

36, Gordon Square,
London, W.C.1
September, 1940.

Dear Colleague,

We are fully conscious of the demands which have been made upon your time and pocket in connection with charitable objects since the commencement of the war, and inevitably these demands will increase. In spite of this, it is with the utmost confidence that we are addressing this appeal to you.

It has been our unhappy lot to witness an ever-increasing amount of unemployment and under-employment within the profession. A number of practices have had to close down, the income from many others has been reduced almost to vanishing point through the destruction of the small animal population, or through the evacuation of large numbers of the civil population with their animals. The Association is actively engaged in an endeavour to

create fresh fields of employment in order to ameliorate the lot of our less fortunate colleagues; we are confident that in this the Association will succeed, though the process is necessarily a slow one.

In spite of the hardships which beset us to-day, it is incumbent upon each and all, more now than ever, to do our utmost to support the Victoria Veterinary Benevolent Fund and the National Veterinary Benevolent and Mutual Defence Society. The demands made upon these societies are increasing, and yet both, unable to make new grants, are actually having to reduce some of those already made to veterinary surgeons, their widows or families, who already are hard put to it to make ends meet. It is our intention, at the Annual General Meeting, to ask members to vote a special sum from the funds of the "National" to one or both of our charities to be used for the benefit of members of this Association.

One of the more important sources of revenue to our benevolent funds is derived from the social activities of the Annual Congress of this Association. Unfortunately, this year, through circumstances outside our control, the Congress has had to be abandoned, and owing to the present uncertain situation it has been found necessary to hold the Annual General Meeting unaccompanied by any social function. The Ladies' Guild has for many years at successive congresses laboured well and unselfishly to raise money for these causes; last year their efforts at the Congress Dance alone brought in nearly £200, this year they will be denied this opportunity so we are addressing this earnest appeal to you to show your appreciation of their previous endeavours by sending us a special donation for these funds, which you would have so willingly and so freely supported had you been present at the Congress. May we also ask you not to lose sight of the fact that had there been a Congress you would have been put to some expense in attending it; this you have now been saved and we would urge you to bear this fact in mind in sending us a contribution to what must be to us the most worthy of all charities—our own veterinary benevolent funds.

Yours sincerely,

H. W. Steele-Bodger,
President.

W. R. Wooldridge,
Hon. Secretary.

H. E. Bywater,
Hon. Treasurer.

The Committee on Traumatic Shock and on Blood Transfusion of the Medical Research Council have issued as M.R.C. War Memorandum No. 1 an excellent summary of methods of treating wound shock (Stationery Office, 4d.). The instructions have been produced in co-operation with the Army Medical Service, and in appendices details are given of the Army blood transfusion outfits and instructions for their use.

ANNOTATION

Vitamins and Muscular Disorders

During the last decade or so there have been striking advances in our knowledge of the actual structure of the vitamins, and many of the "accessory food factors" hitherto designated only by letters of the alphabet have been identified chemically. Thus much is known of the chemical structure of the A and D groups, of B_1 (aneurin, thiamin), of the nicotinic acid, riboflavin and adermin (B_6) fractions of the B_2 complex, of C, E, K and P, and with this knowledge it is becoming more and more possible to produce refined synthetic preparations available for experimental and therapeutic use. Indeed, many of the next steps lie in the identification of chemical structure with pharmacological action, and, as biological activity becomes correlated with certain groupings, it will perhaps be possible to gauge more nearly the exact physiological rôles of the various vitamins, to synthesise homologues and related compounds of varying activities, and to account for species differences in qualitative and quantitative requirements for the various factors.

The clinician, however, must not be content to await too long the findings of the experimental laboratory, although he should be kept informed of progress in such a way as to enable him to formulate reasonable hypotheses of the relationship of vitamin activity to clinical conditions. He has seen, from the classical examples of scurvy, rickets and osteomalacia, beri-beri, pellagra and canine blacktongue, that certain dietary deficiencies may have profound effects upon health and, after all, his criterion is not whether the clinical condition he encounters is an exact counterpart of that produced in the laboratory, but whether or not the condition of the patient may be influenced beneficially by prophylactic or curative therapeutic measures.

As it is becoming realised that the various vitamins play a much wider rôle in metabolism than was once supposed, the possibilities are being explored that they may be of value in the treatment or prevention of hitherto obscure conditions, and recently attempts have been made to utilise certain of them in muscular or neuro-muscular complaints. Deficiency of vitamin E leads to a peculiar flaccid type of paralysis in the suckling rat, while rats weaned normally but thereafter maintained for long periods on E-deficient diets develop a muscular dystrophy and paralysis. The "muscular dystrophy factor" of Goettsch and Pappenheimer[1], required by guinea-pigs and rabbits, has also been identified with vitamin E. In the case of man, various case reports have appeared[2] describing the effect of vitamin E therapy upon muscular dystrophies such as amyotrophic lateral sclerosis and tabes dorsalis; while results are as yet inconclusive, it is only by actual trial that the value of the treatment may be assessed, and it would not be irrational to suggest that, in the case of the foal and other animals, vitamin E therapy be utilised for certain obscure muscular diseases. One school of thought has sought to show that fowl paralysis is in part an E deficiency, but without wishing to enter into a controversial discussion, it is probably correct to say

that the majority of British workers do not regard the field condition which they recognise as fowl paralysis as being related to the syndrome of E deficiency in this species.

Deficiency of adermin, or vitamin B_6, a recently synthesised member of the "B_2 complex," has been associated with the development of convulsions in young animals of various species, *viz.*, the dog[3], fowl[4], pig[5] and rat[6] and, in the case of the last two, the conditions were definitely epileptiform in nature. Pickup[7], in this journal, reported a case in young pigs of epileptiform fits which responded to a change in diet, and it is possible that the condition manifested was related to a B_6 deficiency in the commercial meal employed. Beneficial results in certain muscular dystrophies have been claimed for B_6 in human medicine[2].

It seems that the dietary-deficient pig is especially susceptible to nervous symptoms and to paralysis. Twenty years ago Plimmer[8] found that rapid cures from "rheumatism," which he then believed to be a manifestation of scurvy, resulted when a change from cooked to uncooked diet was made and various nervous or neuro-muscular symptoms, including paralysis of the hindquarters or of all four limbs, were found typical of A deficiency by the Reading workers[9]. More recently, Chick and her co-workers[5] found that a paralysis of the hindquarters developed in "filtrate factor" deficiency—this filtrate factor being an as yet chemically unidentified member of the B_2 complex—and Hughes[10] noted a crippled condition in pigs deprived of riboflavin.

While many of the vitamins mentioned are now or will shortly be available in the pure state commercially, it must be remembered that for animal use a good quality of cod liver oil remains the most ready source of added A and D, that raw liver or brewers' yeast both contain all the B vitamins, that wheat germ oil is a rich source of E (it contains B_6 amongst others), and that members of the *Cruciferae* have been shown to contain high quantities of C. Whatever results may be anticipated or achieved from vitamin therapy, it must also be remembered that sound animal husbandry, with rigid attention to stock, hygiene and dietetics, is the surest safeguard against the many obscure conditions to which this note refers. The main principles of nutrition are now so well known that there is no rational basis for feeding animals on a deficient diet which has to be supplemented by expensive synthetic products.—Alastair N. Worden.

References

[1]Goettsch, M., and Pappenheimer, A. M. (1931.). "Nutritional muscular dystrophy in the guinea-pig and rabbit." *J. exp. Med.* **54.** 145-165.

[2]See *Lancet.* (1940.) ii. pp. 199-200: "Muscular disorders and nutritional deficiency."

[3]Fouts, P. J., Helmer, O. M., Lepkovsky, S., and Jukes, T. H. (1938). "Production of microcytic hypochromic anaemia in puppies on synthetic diet deficient in rat antidermatitis factor (vitmain B_6)." *J. Nutrition.* **16.** 197-207.

[4]Jukes, T. H. (1939.) "Vitamin B_6 deficiency in chicks." *Proc. Soc. exp. Biol., N.Y.* **42.** 180-182.

[5]Chick, H., Macrae, T. F., Martin, A. J. P., and Martin, C. J. (1938). "The water-soluble B-vitamins other than aneurin (vitamin B_1), riboflavin and nicotinic acid required by the pig." *Biochem. J.* **32.** 2207-2224.

[6]Chick, H., El Sadr, M. M., and Worden, A. N. (1940.) "Occurrence of fits of an epileptiform nature in rats maintained for long periods on a diet deprived of vitamin B_6."

[7]Pickup, J. (1939.) "Illness of pigs due to a food deficiency." *Vet. Rec.* **51.** 535-536.

[8]Plimmer, H. A. (1920.) "Note on 'scurvy' in pigs." *Biochem. J.* **14.** 570-571.

[9]Foot, A. S., Golding, J., and Kon, S. K. (1938.) "The vitamin requirements of pigs." (N.I.R.D. Publication No. 462.)

[10]Hughes, E. H. (1939.) "The rôle of riboflavin and other factors of the vitamin-B complex in the nutrition of the pig." *J. Nutrition.* **17.** 527-533.

ABSTRACTS

[ON THE EXPERIMENTAL PRODUCTION OF NEPHRITIS IN RATS BY MEANS OF PARATHYROID HORMONE AND OF VITAMIN D. Chown, B., Lee, M., Teal, J., and Currie, R. (1939.) *J. Path. Bact.* **49.** 273-290. (17 figs. on 4 plates, 1 table, 26 refs.).]

Renal injury may occur in diseases associated with skeletal demineralisation, including hyperparathyroidism, while injury to and calcification of the kidney have also been described as a result of the administration of toxic doses of vitamin D.

Large doses of parathyroid extract, of the order of 10 U.S.P. XI units upwards, were given at varying intervals to young rats of three different laboratory strains. A chronic nephritis usually supervened, and was characterised by primary lesions in the tubules or interstitial tissue; it was often of great severity. The commonest primary lesion was a simple deposition of calcium within the lumen of the tubule and in the adjacent interstitial tissue, but a primary necrosis of the tubular wall, with or without associated calcium deposition, was sometimes encountered. Polycystic kidneys caused by a cystic dilatation of the tubules were also found in many cases. Untreated rats of the same strains, maintained for up to two years under comparable conditions, often showed a mild albuminuria and an occasional renal lesion, but no evidence of an idiopathic or other chronic nephritis.

Vitamin D in daily doses of 1,500 I.U. upwards, with periods of rest, was found to cause an early calcium deposition within the tubules, but only when administered over long periods did it occasionally lead to a chronic nephritis. Renal deposition of calcium in the vitamin D group was increased when 0·1 mg. Ca per gramme body weight (in the form of calcium lactate) was added to the diet.

A. N. W.

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[THE EFFECT OF DIETARY PROTEIN ON THE COURSE OF NEPHROTOXIC NEPHRITIS IN RATS. Farr, L. M., and Smadel, J. E. (1939.) *J. exp. Med.* **70.** 615-628. (9 figs., 1 table, 16 refs.).]

A chronic glomerulonephritis was induced in 48 selected animals of an in-bred strain of hooded rats by three successive daily injections of "anti-rat-kidney serum." On the day following the last injection, the animals were divided into three comparable groups, the first of which was continued on the stock basal diet of 18 per cent. protein, 51 per cent. carbohydrate, 27 per cent. fat and 4 per cent. mineral mixture; the second and third groups were fed respectively on a

high-protein (40 per cent.) and a low-protein (5 per cent.) diet, each of which was isocaloric with the stock diet and contained equal amounts of fat and minerals.

During the first month, the course of the nephritis, as judged by blood and urine analyses, was similar in all three groups. All but two of the rats in the low-protein group then recovered rapidly, although in some of them it was possible to induce an albuminuria by changing to a high-protein diet some months later. In the medium-protein group, seven of the 14 animals which survived the initial phase of the disease apparently died from renal failure. In the high-protein group, a progressive nephritis developed in every case and only two animals were still alive ten and a half months after injection of the anti-kidney serum.

[Although these results appear to have an important bearing on the treatment of nephritis, it must be remembered that the type of nephritis induced by the anti-rat-kidney serum is histopathologically very different from the chronic nephritis in dogs which the practitioner is usually called upon to treat.]

A. N. W.

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[BACTERIOLOGICAL STUDIES OF SPRAY-DRIED MILK POWDER. Nichols, Agnes A. (1939.) *J. Dairy Res.* **10**. 202-230. (14 tables, 20 refs.).]

Spray-dried milk has a very high solubility and can be reconstituted more readily than roller-dried milk; for this reason it is gaining in popularity. It is not, however, subjected to such extensive heat treatment as the roller-dried product, and may therefore be expected to contain a more numerous and varied bacterial flora.

An extensive bacteriological study was made of over 400 samples taken, at different times of the year and at different stages of the day's run, from eight factories in different parts of Great Britain. The samples were reconstituted, and standard plate counts, methylene-blue reduction tests, presumptive coliform tests, and keeping quality tests were carried out; 1 c.c. of the reconstituted milk was also added to litmus milk, while colonies picked off the agar plates used in the counts were also inoculated into litmus milk as a method of isolation.

In general, a very considerable variation in the bacterial quality was found, not only from factory to factory but also from day to day, and it appeared that some deterioration might occur during a single day's run, probably due to the multiplication of thermophilic and thermoduric bacteria during processing. Seasonal variation did not occur in samples from factories where the general standard was high. Great improvement could therefore be obtained in the less satisfactory cases by attention to technique, while the author is of the opinion that grades for spray-dried milk should be introduced.

The predominating flora of the reconstituted product at 37° C. produced an acid reaction and clotted the milk; in almost all cases the responsible bacteria were found to be heat-resistant streptococci, which are further classified and shown to be chiefly *Str. thermophilus* and *Str. durans*. A few *B. subtilis* type organisms causing peptonisation at 37° C., and strains of *B. calidolactis* obtained from plates incubated at 55° C. were also recovered. The number of yeasts and moulds detected was negligible.

A. N. W.

[THE RAPID DETERMINATION OF PEROXIDE VALUES FOR THE FAT IN MILK POWDERS. Smith, J. A. B. (1939.) *J. Dairy Res.* **10**. 294-299.]

The determination of the peroxide value of the fat in milk powders has hitherto been a somewhat tedious procedure, involving the use of an expensive reagent in the peroxide-free ether used for Soxhlet extractions. The method described in the present paper is rapid and simple: 10 g. of milk powder are weighed into a 100 c.c. volumetric flask; 50 c.c. glacial acetic acid are added and the mixture warmed to 35° C. for 5 mins., with shaking at intervals; chloroform from a burette is then added, with shaking, to the 100 c.c. mark, and the mixture rapidly filtered. An aliquot portion of the filtrate is then measured out and, after the addition of 1 c.c. of a saturated solution of potassium iodide and water to 100 c.c., is titrated against standard sodium thiosulphate.

Owing to the extraction of a certain amount of non-fatty material, the titration results are somewhat affected and the values obtained are lower than the true peroxide values by some 10 per cent. The cheapness, ease and rapidity of the method, together with the fact that it may be carried out in ordinary diffused daylight or in artificial light, are held to outweigh this discrepancy for most purposes.

A. N. W.

TREATMENT OF SCABIES

The following method of treatment adopted for human scabies, described in a recent issue of the *British Medical Journal*, will certainly commend itself for trial upon the domestic animals.

"Therapeutics as a whole has advanced very rapidly during the present century, but there are a few subjects that have been little affected by these changes. This is particularly true of skin infestations such as pediculosis and scabies. Prof. P. A. Buxton in a recent discussion remarked that no advance had been made in the technique of delousing since 1918; similarly, the treatment of scabies is still based on sulphur ointment, a remedy which has been used for this purpose for centuries. In most other fields it has been found possible to effect great advances on traditional remedies, and the reason for absence of change in this case is probably lack of research rather than the impossibility of improving established methods. Two American authors, Thomas and Miller, have recently investigated the action of the insecticide rotenone in scabies. Rotenone is one of the active principles of *Derris elliptica*. Crude or partly purified extracts of this plant have found a wide employment in gardens and farms. They are used to kill aphis and in the treatment of warble-fly infestation. The authors used a 1 or 2 per cent. lotion of rotenone on 24 unselected cases of scabies. Cure followed in all cases, and in the majority four applications resulted in complete involution of the lesions in three or four days. The preparation did not stain the bedclothes or underwear and was non-irritating; indeed the patient was not conscious of its presence on the body. This sounds a very great advance on the traditional treatment with sulphur ointment. The authors recommend rotenone for more extended trial in the treatment not only of scabies but also of other dermatoses of parasitic origin."

* * * * *

Acorns, horse chestnuts, and beech mast can be had for the gathering in many districts, and all can contribute usefully to war-time livestock rations. The Ministry of Agriculture's "Growmore" leaflet No. 39 (free on application to the Ministry at Hotel Lindum, Lytham St. Annes, Lancs) gives information about the feeding value of this harvest of the woodlands.

REVIEW

[DISEASES OF THE PIG AND ITS HUSBANDRY. David J. Anthony, M.R.C.V.S., D.V.S.M. (Vict.). Baillière, Tindall & Cox. pp. 272. 48 illus. Price 10s. 6d.]

The pig occupies an ever-increasing share of the time of the country practitioner, and Mr. Anthony's textbook, which is published at what in these times appears a most reasonable price, will be of great value to all of us who concern ourselves with the pig as patient.

Of the fourteen chapters, the first four deal with the breeds, housing, management, breeding and feeding in a most comprehensive way. The eight succeeding chapters cover the diseases of the species. The final chapter deals with poisons.

By way of criticism, we would draw the author's attention to one or two points that might receive attention in the next edition. On page 132, we note that he recommends the hypodermic route for the administration of calcium chloride. We were not aware that the pig was immune from the disastrous results that follow hypodermic use of this compound in other species. We feel that swine fever might receive attention and description more from the angle of a practitioner dealing with a suspected outbreak on behalf of the Ministry of Agriculture, which is the line of approach most of us take. In this connection, we failed to note any mention of the splenic infarct as a diagnostic sign. A detailed account of the mode in which a systematic *post-mortem* examination of the pig should be undertaken, such as was given to some of us at the Yarmouth Congress of the N.V.M.A., would be a welcome addition.

We would urge upon the author and publishers the necessity of providing one or two coloured plates of the skin lesions and bowel lesions of the principal fevers of the pig. How the laboratory decides on its findings from specimens provided by the practitioner is another mystery whose elucidation our author has not attempted to solve !

One final criticism. Will Mr. Anthony let some literary-minded friend run over the script of the next edition ? There is a weariness to the reader in reading sentence after sentence of similar construction with many of the words the same as well. Round about p. 162 let him count the number of times the word " may " occurs in his sentences. Five times in seven lines is too much, and lends a monotonous uncertainty to sentences that might well be more dogmatic in their lay-out.

But let it not be thought that these criticisms are intended to deter the profession from obtaining this work. It is in the main first rate, and may be called indispensable, and with the emendations that will appear in inevitable later editions may well become one of our professional classics.

Harvard University and the American Red Cross will shortly be sending to Great Britain a complete medical unit to study the preventive and clinical problems associated with infectious diseases under war conditions. The unit, which will consist of a hospital and laboratory, will be incorporated in the civil emergency hospital organisation of the Ministry of Health and will be directed by Professor J. E. Gordon of Harvard University.

NOTES AND NEWS

The Editor will be glad to receive items of professional interest for inclusion in these columns.

Diary of Events

Oct. 9th.—Meeting of the Finance and General Purposes Committee, N.V.M.A., at the Randolph Hotel, Beaumont Street, Oxford, 10 a.m.

Meeting of Council, N.V.M.A., at the Randolph Hotel, Beaumont Street, Oxford, 2 p.m. Annual General Meeting of the Ladies' Guild at the same hour and place.

Annual General Meeting of the National Veterinary Medical Association of Great Britain and Ireland, at the Randolph Hotel, Beaumont Street, Oxford, 3 p.m.

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Notice to Members, N.V.M.A.

Meetings at Oxford

The attention of members of the Association is drawn to the following alterations, necessitated by the emergency situation, which have been made in the arrangements, as notified with the Annual Report, for holding the Annual General Meeting. The meeting will be held on Wednesday, April 9th, at the Randolph Hotel, Beaumont Street, Oxford, at 3 p.m. The members of the Finance and General Purposes Committee will meet at the same venue in the morning, when all members of other committees have permission to attend. A meeting of the Council of the Association will precede the Annual General Meeting. It is hoped that as many members as possible will attend an informal luncheon at the Randolph Hotel, Oxford, on that day: all who can do so are requested to communicate with the General Secretary at the address given below.

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Association Correspondence

The General Secretary of the N.V.M.A., Mr. F. Knight, has taken up his residence on premises of the President, Mr. H. W. Steele-Bodger, at 10, Bore Street, Lichfield. In order, however, to facilitate consultation with the President, where necessary, all correspondence for the General Secretary should be addressed to him **c/o H. W. Steele-Bodger, Esq., M.R.C.V.S., 63, Lichfield Road, Sutton Coldfield, Warwickshire (Tel.: Sutton Coldfield 1608 and 1609).** This arrangement comes into operation on Monday next. All correspondence for the Editorial Representative, N.V.M.A. (including literary matter and advertisements) should be addressed to him as heretofore at 36, Gordon Square, London, W.C.1.

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PERSONAL

Birth.—Cockrill.—On September 17th, to Eudora (*née* Sime), M.R.C.V.S., wife of W. Ross Cockrill, M.R.C.V.S., " Maes-yr-awel," College Road, Carmarthen—a daughter.

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Marriages. — Barr — Atkinson. — At St. Michael's Church, Barton, on September 19th, 1940, by the Rev. E. Hudson; John Barr, M.R.C.V.S., only son of the late Thos. Barr and Mrs. Barr, Crown Square, Penrith, to Janet Howe, only daughter of the late James Atkinson and Mrs. Atkinson, Kirkbarrow, Tirril, Penrith.

CAMPBELL—WALLACE.—September 12th, 1940, at 1st Kilraughts Presbyterian Church, by Rev. F. A. Robinson, B.A., Donald William Campbell, M.R.C.V.S., younger son of Francis and Mrs. Campbell, Evanton, Ross-shire, to Margaret Mathews, fourth daughter of James Moore and Mrs. Wallace, Broughanore, Culcrum P.O., Co. Antrim.

CARNEGIE—BOON.—On September 19th, at the Parish Church, Kendal, William K. Carnegie, B.Sc., M.R.C.V.S., Brampton, Cumberland, to Marjorie Webster, only daughter of Mr. and Mrs. F. W. Boon, Eller Lea, Kendal.

FERGUSON—MAXWELL.—The marriage between Hugh Noel Ferguson, M.R.C.V.S., and Elinor R. Maxwell took place on September 25th at Kirkcubbin Presbyterian Church, Co. Down, Northern Ireland.

HILL—LEWIS.—On September 7th, at St. Andrew's, Dinas Powis, Fergus John Hill, M.R.C.V.S., D.V.S.M., to Dora Mary, elder daughter of Mr. and Mrs. W. E. Lewis, Mountside, Dinas Powis, Glamorgan.

LEASK—BENNETT.—At St. Michael's Parish Church, Macclesfield, Cheshire, on September 18th, 1940, by the Rev. J. E. D. Devereux, M.A., Douglas Ewart Leask, M.R.C.V.S., younger son of Mr. W. S. Leask, Registrar, 15, Low Street, Banff, to Winifred May Bennett, elder daughter of Mr. and Mrs. John Bennett, Belmont, Oxford Road, Macclesfield.

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GENERAL OBITUARY

MR. JOHN ANDERSON

Old students of the School of Veterinary Science at Liverpool University will learn with regret of the passing of Mr. John Anderson. He had been in failing health for some weeks and he passed away in hospital on September 12th. The funeral, which was private, took place at Anfield Crematorium on Saturday, September 14th, 1940. He leaves a widow and one daughter.

A native of Elgin, Mr. Anderson came to Liverpool from the New Veterinary College, Edinburgh, when the University School was founded in 1904. He was Professor Share Jones's Senior Attendant in the Department of Veterinary Anatomy, and both in the dissecting room and outside he came into intimate contact with veterinary students.

John, as he was affectionately known to staff and students, was a loyal servant of the School, and was popular with all. He was also keenly interested in matters which concerned the veterinary profession and he was jealous of its traditions. He retired in 1939 and at a gathering of staff and students, when presentations were made to him, he was proud to boast that he had not been absent on any day throughout the period of his association with the University.

Many old graduates of Liverpool will feel the loss of one who was guide, philosopher and friend to them in those impressionable days of their student life.

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VETERINARY COUNCIL (IRELAND)

A meeting of the Veterinary Council of Ireland took place on Wednesday, September 18th, presided over by Mr. A. A. Donnelly, M.R.C.V.S.

The following members were present: Professor O'Connor, M.R.C.V.S., Professor W. Kearney, M.R.C.V.S., Mr. P. F. Dolan, M.R.C.V.S., and Mr. James Dodd, M.R.C.V.S.

Messrs. A. A. Donnelly, M.R.C.V.S., P. F. Dolan, M.R.C.V.S., D.V.S.M., and Mr. James Dodd, M.R.C.V.S., were nominated to the Standing Committee of the Cultural and Education Panel in accordance with the provisions of the Seanad Electoral (Panel Members) Act, 1940.

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Notification is made in our advertisement columns that Dr. Tom Hare, with the assistance of Mr. A. B. Orr, has opened his own laboratories, for service on behalf of members of the veterinary profession, at 529a, Finchley Road, London, N.W.6.

CHEMOTHERAPEUTIC THEORY

"The process whereby sulphonamide compounds restrain bacterial growth in the body remains in dispute," commented the *British Medical Journal* recently. "Their action on bacteria *in vitro* is too feeble to support the idea of a direct antiseptic action in the ordinary sense, and it has seemed reasonable to postulate some more subtle interference with the activities of the bacterial cell; indeed, this is not pure assumption, since there is evidence, for example, that sulphanilamide can prevent bacterial utilisation of serum proteins as sources of nutriment. Another kind of explanation which has been repeatedly offered, particularly by French authors, is conversion in the body into another product with much greater direct anti-bacterial activity. Often propounded only in the vaguest general terms, this idea was given precise form by R. L. Mayer[1], who showed that *p*-hydroxylaminobenzene sulphonamide is actively bactericidal, and argued that the formation of this substance from sulphanilamide in the body is feasible. This conversion could not take place in the absence of oxygen, and it may therefore be significant, as P. A. Shaffer[2] has recently pointed out, that sulphanilamide and sulphapyridine are both deprived of all bacteriostatic action under anaerobic conditions. Shaffer is among several workers who have studied the changes produced in these drugs by various physical and chemical agencies, and by the use of oxidising agents he has obtained unstable products, the exact constitution of which is so far undetermined, which possess high oxidising potentials. The activity of these substances as indicated by potential measurements is confirmed by their chemical reactions, and among their properties is that of rapidly converting oxyhaemoglobin into methaemoglobin. This introduces another aspect of the subject, for among the results of sulphonamide medication which remain to be fully explained is the constant occurrence of methaemoglobinaemia, the usual basis of the familiar cyanosis. The formation of an active oxidizing agent from the drug would fully account for this change in the blood, and argument from this angle therefore makes it seem all the more probable that some such product is formed in the body. It must nevertheless be remembered that in all the work so far quoted there is no direct proof of this; still less is the mechanism of its formation known, although Shaffer offers a plausible tentative explanation of it. It would therefore be of the highest significance if sulphanilamide could be shown to acquire any of the properties of these oxidation products under the influence not of chemical reagents but of living tissue.

"In the work of J. S. Harris[3] this now appears to have been done. Slices of normal tissue from freshly killed animals were incubated for two hours with suspensions of red cells in Ringer's solution containing various concentrations of sulphanilamide. The red cells were then spun out, washed, and lysed with saponin, and the concentration of methaemoglobin in the resulting solution was measured by spectrophotometer. Whereas in mixtures containing no tissue the amount of methaemoglobin formed never exceeded 1 per cent. of the total haemoglobin, the presence of rat liver—the most active tissue tried—caused the conversion of 43 per cent. when an optimum concentration of sulphanilamide, which was found to be 0·1 per cent., was also present. The liver of other animals had the same effect in somewhat less degrees; mouse kidney was also active and rat kidney less so; muscle, spleen, and brain had little or no effect. The combination in one mixture of all the elements concerned in this process is not necessary; if a sulphanilamide solution containing liver slices is incubated and then cleared by centrifugation, the supernatant fluid will cause methaemoglobin formation when added to red cells. Nor is the intact red cell necessary; the product of interaction between tissue

(1) *Bull. Acad. Méd.*, Paris. (1937.) **117**. 727.
(2) *Science*. (1939.) **89**. 547.
(3) *J. clin. Invest.* (1939.) **18**. 521.

and sulphanilamide will convert haemoglobin in solution into methaemoglobin, although the results here are less conclusive owing to the more extensive conversion taking place in controls with no drug. Comparison between these results and those obtained by Harris and H. O. Michel[4] in a clincal study of methaemoglobinaemia due to sulphanilamide is said to show a correspondence between them in that the influence of sulphanilamide concentration, and even of the concentration of haemoglobin itself, on the amount of methaemoglobin formed is of the same nature in both. No attempt has been made to identify chemically the oxidising agent which thus appears to be formed from sulphanilamide in the presence of certain tissues. The fluid in which this reaction occurs remains colourless, and the agent, whatever it may be, is therefore not the same as Shaffer's oxidation products, which are cherry-red, or the blue substance formed when sulphanilamide in solution is subjected to ultra-violet light.

"Harris's observations do not support the hypothesis of Main, Shinn, and Mellon that the therapeutic action of sulphanilamide depends on anticatalase activity; he gives three reasons, each sufficient in itself, why the substance formed in his experiments cannot be supposed to act in this way. Mayer's original hypothesis and the subsequent observations quoted here seem to hang plausibly together, but there are several experimental steps to be taken before the whole of this explanation of the behaviour of sulphonamide compounds in the body can be accepted. If it should prove to be correct it will be of more than academic interest. As Shaffer points out, if therapeutic activity depends on the oxidation-reduction potential of compounds formed from the drug in the body this fact affords both a basis for relating molecular structure to activity and a comparatively simple method of *in vitro* test of probable therapeutic usefulness."

(4) Ibid. 507.

* * * * *

SILAGE

Observing that if there was enough rain within the next few weeks to make the grass grow we should have thousands of tons of silage before next winter and that farmers who had never made silage before were ready to start then, the *Lancet* asked, in a late August issue, "Why this sudden interest?" and proceeded: "Silage is nothing new, for the process, which may be described as the pickling or preserving in a succulent condition of various farm crops, was introduced into this country 50 years ago. But the type of crop which could then be successfully used was limited. It was also considered necessary to employ large concrete containers called tower silos which few farmers could afford, and the resulting silage varied widely in feeding value according to the farmers' knowledge of the process. To-day the limitations have largely been overcome, and owing to the difficulty of importing feeding-stuffs every farmer is trying to ensure a supply from his own farm. The Ministry of Agriculture is arranging demonstrations to popularise the process, and apart from green crops grown specifically for silage they are urging that all surplus grass and other crops should be ensiled against winter scarcity.

"The principles of silage-making are to cut the crop in a green state and then to handle it so as to curtail leaf respiration and avoid the production of temperatures over 120° F. which would lead to the loss of nutrient material. This is achieved by packing the greenstuff tightly in a closed container or silo which rations the supply of air. As respiration proceeds and the mass warms up, enzymes break down both proteins and carbohydrates. Ultimately, on the death of the plant cells, micro-organisms become active and various organic acids are formed which act as preservatives. The most important of these acids is lactic acid, formed from the sugar of the crop. Unless the crop contains enough starch or sugar undesirable acids such as butyric acid may be formed, giving the silage an evil smell. For this reason until recently it was not thought possible to ensile crops, such as young leafy grass, which were rich in protein but poor in starch or sugar. This difficulty has been overcome by the simple expedient of adding sugar in the form of molasses—whey was first tried—and in this way the farmer can make silage from protein-rich material and find himself largely independent of imported cake. Yet another incentive to silage-making is that it is no longer necessary to incur high capital expenditure in building a large fixed tower silo. Cheap portable silos are now available in sections of wood or wire mesh lined with special paper. Again, silage can be made at odd times when the weather is unsuitable for other farm operations.

Well-made silage is a succulent yellowish material with a slightly acid taste and a faint but pleasant smell. Its food value depends on how and from what it is made. Ordinarily silage, on a dry-matter basis, has a food value equal to hay, but molassed silage made from young leafy grass may contain twice the protein of hay and rival a cake or other concentrated food. Silage and hay are not in fact interchangeable in stock dietetics, but are rather complementary, the silage adding succulency to the animals' diet. Normally silage contains more carotene than hay, and in late January, when milk and butter lose their colour and with it much of their vitamin-A content, the feeding of good silage will ensure that the consumer gets a milk rich in vitamin at a time when other protective foods are scarce. Moreover, though Hitler may try to fire our haystacks he will find it difficult to destroy silage with his incendiary bombs."

* * * * *

THE SOYA BEAN IN DIETETICS

"The soya bean has been described as vitally important to Germany from the nutritional, the economic, and the military standpoint," states the *British Medical Journal*, and continues: "A correspondent in the *Times* described a few weeks ago[1] how these 'Nazi food pills' were playing a dominant part in supplying the protein missing in a diet in which foodstuffs of animal origin were likely to be deficient. A flour from the soya bean containing 40 to 45 per cent. of protein has been made and is incorporated in soups, sausages, bread, biscuits, macaroni, etc., in such a way that flavour is unimpaired and the food value greatly increased. From the military aspect it has been openly boasted in Germany that without soya flour the rapid advances of mechanised units, cut off by distance from food supplies, would not have been possible. Even allowing for the flights of imagination which colour reports from the Reich, it is clear that the possibilities of the soya bean require more consideration than is usually given to them by dietetic experts in this country. A recent account[2] by Dr. Helen Mackay of the use of soya bean flour in infant feeding is therefore of more than usual interest. The flour used contained over 40 per cent. of protein, 20 per cent. fat, and 20 per cent. carbohydrate. It contained about a quarter as much calcium, half as much phosphorus, and about eight times as much iron as dried cow's milk. For the investigation the soya flour was mixed with equal parts by weight of a full-cream dried milk. When reconstituted with the usual 'one in eight' dilution this mixture gave a feed with a calorie value equal to that of liquid cow's milk. Babies were given this milk, while a control group had dried milk with added iron. All babies received orange juice and some form of vitamin D, and from six months of age all began mixed feeding. Babies accepted for the final comparison of results numbered 150. It was found that those receiving the soya flour milk gained on the average a little more slowly in weight than the controls, but the difference was less than 2 ounces a month. The haemoglobin levels of the soya flour group remained very slightly below that of the controls, who had extra iron, and the total morbidity rates were almost the same in the two groups. There was, however, a greater tendency to

(1) *Times*, April 23rd, 1940.
(2) *Arch. Dis. Childh.* (1940.) 15. 1.

loose stools when the soya bean flour was used, and apparently it was not so well tolerated as full-cream dried milk and must be introduced more gradually. Calcification of the bones was as good in the soya bean group as in the controls. Dr. Mackay has proved that in the mixture described soya bean flour can play a part in infant feeding. From the economic aspect it is pointed out that the relative cheapness of soya bean flour should make it possible to market a milk plus flour powder at a price much lower than that of full-cream dried milk. Writing at a time when the possible stringencies of war economy were not fully realised, Dr. Mackay concluded that 'owing to its relative cheapness it [the soya bean flour-milk mixture] merits an extended trial in countries where, for economic reasons, a satisfactory substitute for breast milk has hitherto been unobtainable.' The story of the 'food pills' in Germany suggests an even wider application of soya flour. Infant feeding provides a delicate test for a substitute material, and out of such a test in Dr. Mackay's hands the soya bean emerges with great credit. The main sources of the plant are Manchuria, the United States of America, the Balkans, and South Russia. It is to be hoped that someone in the Ministry of Food is interested in the soya bean."

* * * * *

ANDROGENS AND CANCER

"Oestrogenic hormones, beyond a doubt, play an essential role in the induction of mammary cancer in mice," stated the *British Medical Journal* in an annotation (published in the issue of July 6th) which continues: "This being demonstrated, it is of evident importance to inquire if the action of the oestrogens can be annulled by the administration of androgens. Nathanson and Andervont[1] and Lacassagne and Raynaud[2] have shown that androgens exert an antagonistic action. Administration of testosterone propionate greatly reduces the incidence of mammary cancer in mice which are prone to the disease. Lacassagne[3] concludes that testosterone prevents mammary cancer by causing an inhibition of pituitary secretion and, secondarily, a suppression of ovarian secretion. The mice receiving testosterone are comparable, therefore, with ovariectomised mice, in which, as L. Loeb found many years ago, the incidence of mammary cancer is low. These experiments are concerned with the initiation of mammary cancer. Testosterone does not stop the progress of a mammary cancer which is in being when the hormone is first administered; it has a prophylactic but no therapeutic effect in mice. There is evidence that growth of transplantable benign fibro-adenoma of the breast in rats is controlled to some extent by hormones, and that these also have some effect on fibro-adenoma of the breast in women. The aggravation of cancer of the breast during pregnancy and lactation suggests that the growth of breast cancer in women can be influenced by hormones. Ovariectomy was at one time a vaunted treatment for breast cancer and still has its adherents, but it is undoubtedly safer to remove the tumour and leave the ovaries than to remove the ovaries and leave the tumour. In some patients, ovariectomy is beneficial, but is in no way a cure. No greater success can be expected to result from suppressing ovarian function by the administration of testosterone to patients who already suffer from mammary cancer. If androgens retard the growth of tumours it is to be expected that their action will be exerted most conspicuously and perhaps exclusively on tumours of the reproductive organs. The failure of testosterone to influence the growth of mammary cancer in mice does not encourage the expectation that tumours of other types will respond to similar treatment. Murlin and others have described an adverse effect of androgens on the growth and metastasis of the Brown-Pearce rabbit tumour, which originated as an epithelioma of the skin of the scrotum. The results are not impressive. By using a transplantable tumour, and, moreover, one which often disappears without interference, Murlin and his colleagues have complicated the issue. A transplanted tumour is alien to its host and is exposed to to hostile reactions which are not exerted against spontaneous tumours. Many animals have rid themselves of transplantable tumours without aid or with the help of treatments which fail to impede the course of human cancer for a day."

(1) *Proc. Soc. exp. Biol.* N.Y. (1939.) **40**. 421.
(2) *C. r. Soc. Biol.*, Paris. 1939. **131**. 586.
(3) *Bull. Cancer.* (1939.) **28**. 951.

* * * * *

REACTIONS BETWEEN OVARY AND PITUITARY

"It has been known for many years that hypophysectomy leads to ovarian atrophy, and that this can be prevented by the implantation of pituitary tissue," states the *British Medical Journal* in a recent annotation. "After this discovery gonadotrophic extracts were prepared from the pituitary and other potent sources—urine from the pregnant woman and serum from the pregnant mare. The earlier testing of such extracts was carried out largely on normal rats or mice. The observation of Selye, Collip, and Thompson[1] that extracts of pregnancy urine (chorionic gonadotrophin) when injected into immature hypophysectomised rats produced only luteinisation, although provoking both follicle stimulation and luteinisation when injected into normal immature rats, showed that the pituitary of the test animal seriously interfered with the results. Noble, Rowlands, Warwick, and Williams,[2] comparing the effects of numerous gonadotrophic extracts on normal and hypophysectomised immature female rats, found that in all cases the quantitative response in the latter was less than in the former. The qualitative differences were in some cases, however, even more striking. Whereas all the extracts gave rise to follicle stimulation and luteinisation in the normal rats, those from the urine and serum of pregnant women and from the ox hypophysis were purely luteinising, and those from certain human glands purely follicle stimulating, in the hypophysectomised rats. Extracts from the pituitaries of sheep, horse, and pig, and from the serum of the pregnant mare, had both activities even in the operated animals. All this shows that the injection of follicle-stimulating gonadotrophin provokes the secretion of luteinising gonadotrophin, and *vice versa*. It is difficult to determine whether these responses are elicited by the injected materials themselves or by the secretions of the ovary in its follicular and luteal phases. Large doses of oestrogen have an inhibitory effect on the pituitary, as is shown by Ruth Deanesly,[3] who has observed that implantation of oestrone or oestradiol tablets in male rats or mice leads to degeneration of the gonads and accessory sex organs. This is attributed to hypophyseal depression, since the growth rate is also diminished. Similar results have been obtained in rats of both sexes by Noble,[4] using stilboestrol tablets. He also demonstrated that the gonadotrophin content of the pituitary of the animals so treated was diminished. The qualitative aspects of this diminution are not yet established, nor are the similar aspects of the diminution produced by progesterone. It is apparent that the relations between ovary and pituitary are complex, and that complete replacement therapy in hypophysectomised animals will present great difficulty until the constituents of the gonadatrophic complex are separated in a physiologically pure state. The difficulty is exemplified in the work of Liu and Noble,[5] who find that, while the loss in ovarian weight from hypophysectomy can be arrested by daily injection of extract of pregnant mare's serum, the effect does not last and

(1) *Endocrinol.* (1933.) **17**. 494.
(2) *J. Endocrinol.* (1939.) **1**. 22.
(3) *Ibid.*, 36.
(4) *Lancet.* (1938.) **2**. 192.
(5) *J. Endocrinol.* (1939.) **1**. 15.

regression sets in after a few weeks in spite of continued injection. During the injection period the animals are continuously in oestrus, and although mating occurs pregnancy does not result. Injections of chorionic gonadotrophin had similar effects, though the regression in ovarian weight set in earlier and oestrus only persisted for a few days."

* * * * *

A certain journal, in its report of an air raid in a country area said that a farmer there lost "a valuable Suffolk colt and some Lincoln cows." The report was submitted to the censor. When it returned the words "Suffolk" and "Lincoln" were erased. Presumably the censor thinks that Jerseys can only be farmed in Jersey, and that Suffolk horses never stray over the county border.—*Farmers' Weekly.*

CORRESPONDENCE

Letters to the Editor should reach the Office not later than by the first post on Tuesday morning for insertion in following Saturday's issue.

The views expressed in letters addressed to the Editor represent the personal view of the writer only and must not be taken as expressing the opinion or having received the approval of the N.V.M.A.

* * * * *

FUND FOR THE RED CROSS AND VETERINARY CHARITIES

To the Editor of the Veterinary Record

Sir,—The response to my recent letter appealing for medals has been very disappointing—only two contributions have been received instead of the dozens for which I had hoped. I should be grateful if you can spare room in your columns for the list of contributors to date in the hope that it may stimulate others to give when repeated appeals in your columns have failed.

The list is as follows:—

	£	s.	d.
Miss M. L. Bingham	3	6	0
E. Cotchin	2	8	0
Miss M. J. Farman	1	4	0
R. E. Glover	3	1	0
R. Lovell	0	5	0
A. E. G. Markham	1	10	0
C. W. Ottaway	0	10	6
N. J. Scorgie	1	1	0
D. W. B. Taylor	4	1	0
E. G. White	2	8	0
J. G. Wright	0	10	0
"A Glasgow Graduate"	1	1	0
Total	£21	5	6

Of this amount the sum of £5 15s. 6d. is to be given to the Victoria Veterinary Benevolent Fund and the remainder to the Red Cross. This distribution is at the wish of individual donors.

The need is far greater than when this appeal was launched only a few weeks ago and further contributions would be welcome. As pointed out recently in the correspondence columns of *The Times*, appeals for Spitfires and bombers should not be allowed to interfere with the flow of funds to the Red Cross, the needs of which are greater than ever.

Yours faithfully,
E. G. White.

Research Institute in Animal Pathology,
Royal Veterinary College,
"The Beeches,"
Streatley,
Berks.

September 19th, 1940.

DISEASES OF ANIMALS ACTS, 1894 to 1937, and AGRICULTURE ACT of 1937 (PART IV)

Summary of Returns of Confirmed Outbreaks of Scheduled (Notifiable) Diseases

Period	Anthrax.		Foot-and-Mouth Disease.		Parasitic§ Mange.		Sheep Scab.	Swine Fever.	
	Outbreaks confirmed.	Animals attacked.	Outbreaks confirmed.	Animals slaughtered as diseased or exposed to infection.	Outbreaks confirmed.	Animals attacked.	Outbreaks confirmed.	Outbreaks confirmed.	Swine slaughtered.
	No.	No.	No.	No.	No.	No.	No.	No.	No.
Period 16th to 31st Aug., 1940	28	37	—	—	—	—	2	256	215
Corresponding period in ...									
1939	20	25	19	1,979	—	—	2	133	104
1938	19	24	3	203	—	2	9	33	40
1937	18	23	—	—	3	4	6	26	22
Total 1st January to 31st Aug. 1940	367	424	24	3,665	15	30	126	3,020	2,114
Corresponding period in									
1939	547	600	63	7,631	31	71	166	1,581	1,177
1938	580	937	160	20,464	42	72	116	443	342
1937	490	596	19	4,473	91	129	139	748	460

Note.—The figures for the current year are approximate only. § Excluding outbreaks in Army Horses.

Tuberculosis (Attested Herds) Schemes

The number of Attested Herds, *i.e.* herds officially certified as free from tuberculosis, as at 31st August, 1940, was as follows:— England 3,795 ... Wales 9,364 ... Scotland 3,127 ... Total (Great Britain) 16,286

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[No. 40]

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